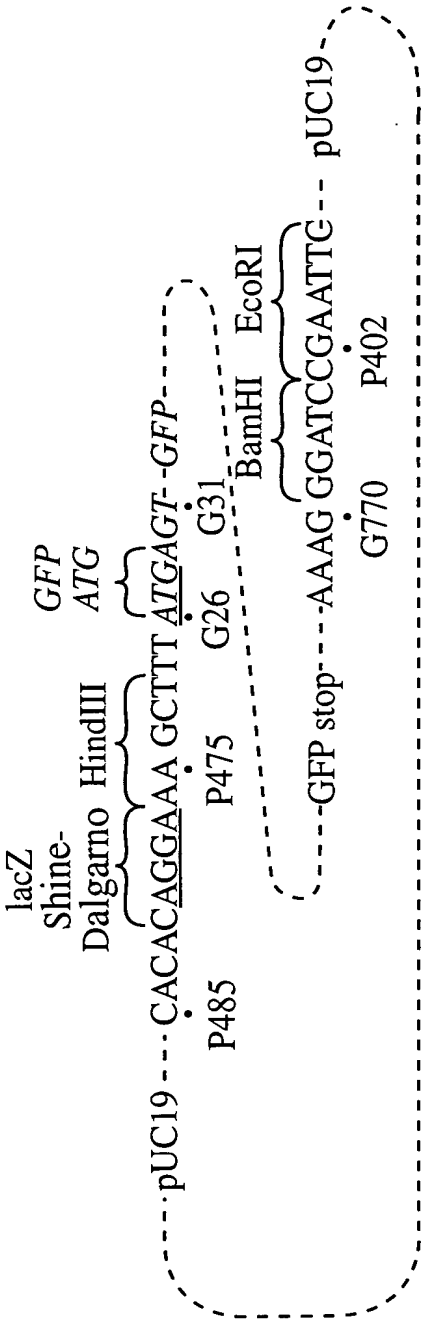
lacZ
Shine-
Dalgarno
HindIII
GFP
ATG
pUC19
CACACAGGAAA GCTTT ATGAGT -GFP
P485
P475
G26
G31
BamHI
EcoRI
-GFP stop
AAAG GGATCCGAATTC
pUC19
G770
P402

FIG. 1

DNA and Predicted primary amino acid sequence of GFP (Hind3-EcoR1 fragment).

5' - AAGCTTT

ATG	AGT	AAA	GGA	GAA	GAA	CTT	TTC	ACT	GGA	GTT	GTC	CCA	ATT	CTT	GTT	GAA	TTA	GAT	CGC
MET	SER	LYS	GLY	GLU	GLU	LEU	PHE	THR	GLY	VAL	VAL	PRO	ILE	LEU	VAL	GLU	LEU	ASP	GLY
GAT	GTT	AAT	GGG	CAA	AAA	TTC	TCT	GTT	AGT	GGA	GAG	GGT	GAA	GGT	GAT	GCA	ACA	TAC	GGA
ASP	VAL	ASN	GLY	GLN	LYS	PHE	SER	VAL	SER	GLY	GLU	GLY	GLU	GLY	ASP	ALA	THR	TYR	GLY
AAA	CTT	ACC	CTT	AAA	TTT	ATT	TGC	ACT	ACT	GGG	AAG	CTA	CCT	GTT	CCA	TGG	CCA	ACG	CTT
LYS	LEU	THR	LEU	LYS	PHE	ILE	CYS	THR	THR	GLY	LYS	LEU	PRO	VAL	PRO	TRP	PRO	THR	LEU
GTC	ACT	ACT	TTC	TCT	TAT	GGT	GTT	CAA	TGC	TTT	TCA	AGA	TAC	CCA	GAT	CAT	ATG	AAA	CAG
VAL	THR	THR	PHE	SER	TYR	GLY	VAL	GLN	CYS	PHE	SER	ARG	TYR	PRO	ASP	HIS	MET	LYS	GLN
CAT	GAC	TTT	TTC	AAG	AGT	GCC	ATG	CCC	GAA	GGT	TAT	GTA	CAG	GAA	AGA	ACT	ATA	TTT	TAC
HIS	ASP	PHE	PHE	LYS	SER	ALA	MET	PRO	GLU	GLY	TYR	VAL	GLN	GLU	ARG	THR	ILE	PHE	TYR
AAA	GAT	GAC	GGG	AAC	TAC	AAG	ACA	CGT	GCT	GAA	GTC	AAG	TTT	GAA	GGT	GAT	ACC	CTT	GTT
LYS	ASP	ASP	GLY	ASN	TYR	LYS	THR	ARG	ALA	GLU	VAL	LYS	PHE	GLU	GLY	ASP	THR	LEU	VAL
AAT	AGA	ATC	GAG	TTA	AAA	GGT	ATT	GAT	TTT	AAA	GAA	GAT	GGA	AAC	ATT	CTT	GGA	CAC	AAA
ASN	ARG	ILE	GLU	LEU	LYS	GLY	ILE	ASP	PHE	LYS	GLU	ASP	GLY	ASN	ILE	LEU	GLY	HIS	LYS
ATG	GAA	TAC	AAT	TAT	AAC	TCA	CAC	AAT	GTA	TAC	ATC	ATG	GCA	GAC	AAA	CCA	AAG	AAT	GGA
MET	GLU	TYR	ASN	TYR	ASN	SER	HIS	ASN	VAL	TYR	ILE	MET	ALA	ASP	LYS	PRO	LYS	ASN	GLY
ATC	AAA	GTT	AAC	TTC	AAA	ATT	AGA	CAC	AAC	ATT	AAA	GAT	GGA	AGC	GTT	CAA	TTA	GCA	GAC
ILE	LYS	VAL	ASN	PHE	LYS	ILE	ARG	HIS	ASN	ILE	LYS	ASP	GLY	SER	VAL	GLN	LEU	ALA	ASP
CAT	TAT	CAA	CAA	AAT	ACT	CCA	ATT	GGC	GAT	GGC	CCT	GTC	CTT	TTA	CCA	GAC	AAC	CAT	TAC
HIS	TYR	GLN	GLN	ASN	THR	PRO	ILE	GLY	ASP	GLY	PRO	VAL	LEU	LEU	PRO	ASP	ASN	HIS	TYR
CTG	TCC	ACG	CAA	TCT	GCC	CTT	TCC	AAA	GAT	CCC	AAC	GAA	AAG	AGA	GAT	CAC	ATG	ATC	CTT
LEU	SER	THR	GLN	SER	ALA	LEU	SER	LYS	ASP	PRO	ASN	GLU	LYS	ARG	ASP	HIS	MET	ILE	LEU
CTT	GAG	TTT	GTA	ACA	GCT	GCT	GGG	ATT	ACA	CAT	GGC	ATG	GAT	GAA	CTA	TAC	AAA	TAA	
LEU	GLU	PHE	VAL	THR	ALA	ALA	GLY	ILE	THR	HIS	GLY	MET	ASP	GLU	LEU	TYR	LYS		

ATGTCCAGACTTCCAATTGACACTAAAGGGATCCGAATTC – 3'

Fig. 2a

Nucleotide sequence (764bp) of GFP (Hind3-EcoR1 fragment)

AAGCTTTATGAGTAAAGGAGAAGAACTTTTCACTGGAGTT
GTCCCAATTCTTGTTGAATTAGATGGCGATGTTAATGGGC
AAAAATTCTCTGTTAGTGGAGAGGGTGAAGGTGATGCAAC
ATACGGAAAACTTACCCTTAAATTTATTTGCACTACTGGG
AAGCTACCTGTTCCATGGCCAACGCTTGTCACTACTTTCT
CTTATGGTGTTCAATGCTTTTCAAGATACCCAGATCATAT
GAAACAGCATGACTTTTTCAAGAGTGCCATGCCCGAAGGT
TATGTACAGGAAAGAACTATATTTTACAAAGATGACGGGA
ACTACAAGACACGTGCTGAAGTCAAGTTTGAAGGTGATAC
CCTTGTTAATAGAATCGAGTTAAAAGGTATTGATTTTAAA
GAAGATGGAAACATTCTTGGACACAAAATGGAATACAACT
ATAACTCACATAATGTATACATCATGGCAGACAAACCAAA
GAATGGCATCAAAGTTAACTTCAAAATTAGACACAACATT
AAAGATGGAAGCGTTCAATTAGCAGACCATTATCAACAAA
ATACTCCAATTGGCGATGGCCCTGTCCTTTTACCAGACAA
CCATTACCTGTCCACGCAATCTGCCCTTTCCAAAGATCCC
AACGAAAAGAGAGATCACATGATCCTTCTTGAGTTTGTAA
CAGCTGCTGGGATTACACATGGCATGGATGAACTATACAA
ATAAATGTCCAGACTTCCAATTGACACTAAAGGGATCCGA
ATTC

Fig. 2b

DNA and predicted primary amino acid sequence of F64L-Y66H-GFP (Hind3-EcoR1 fragment).

5' - AAGCTTT

ATG	AGT	AAA	GGA	GAA	GAA	CTT	TTC	ACT	GGA	GTT	GTC	CCA	ATT	CTT	GTT	GAA	TTA	GAT	GGC
MET	SER	LYS	GLY	GLU	GLU	LEU	PHE	THR	GLY	VAL	VAL	PRO	ILE	LEU	VAL	GLU	LEU	ASP	GLY
GAT	GTT	AAT	GGG	CAA	AAA	TTC	TCC	GTT	AGT	GGA	GAG	GGT	GAA	GGT	GAT	GCA	ACA	TAC	GGA
ASP	VAL	ASN	GLY	GLN	LYS	PHE	SER	VAL	SER	GLY	GLU	GLY	GLU	GLY	ASP	ALA	THR	TYR	GLY
AAA	CTT	ACC	CTT	AAA	TTT	ATT	TGC	ACT	ACT	GGG	AAG	CTA	CCT	GTT	CCA	TGG	CCA	ACG	CTT
LYS	LEU	THR	LEU	LYS	PHE	ILE	CYS	THR	THR	GLY	LYS	LEU	PRO	VAL	PRO	TRP	PRO	THR	LEU
GTC	ACT	ACT	CTC	TCT	CAT	GGT	GTT	CAA	TGC	TTT	TCT	AGA	TAC	CCA	GAT	CAT	ATG	AAA	CAG
VAL	THR	THR	LEU	SER	HIS	GLY	VAL	GLN	CYS	PHE	SER	ARG	TYR	PRO	ASP	HIS	MET	LYS	GLN
CAT	GAC	TTT	TTC	AAG	AGT	GCC	ATG	CCC	GAA	GGT	TAT	GTA	CAG	GAA	AGA	ACT	ATA	TTT	TAC
HIS	ASP	PHE	PHE	LYS	SER	ALA	MET	PRO	GLU	GLY	TYR	VAL	GLN	GLU	ARG	THR	ILE	PHE	TYR
AAA	GAT	GAC	GGG	AAC	TAC	AAG	ACA	CGT	GCT	GAA	GTC	AAG	TTT	GAA	GGT	GAT	ACC	CTT	GTT
LYS	ASP	ASP	GLY	ASN	TYR	LYS	THR	ARG	ALA	GLU	VAL	LYS	PHE	GLU	GLY	ASP	THR	LEU	VAL
AAT	AGA	ATC	GAG	TTA	AAA	GGT	ATT	GAT	TTT	AAA	GAA	GAT	GGA	AAC	ATT	CTT	GGA	CAC	AAA
ASN	ARG	ILE	GLU	LEU	LYS	GLY	ILE	ASP	PHE	LYS	GLU	ASP	GLY	ASN	ILE	LEU	GLY	HIS	LYS
ATG	GAA	TAC	AAT	TAT	AAC	TCA	CAT	AAT	GTA	TAC	ATC	ATG	GCA	GAC	AAA	CCA	AAG	AAT	GGC
MET	GLU	TYR	ASN	TYR	ASN	SER	HIS	ASN	VAL	TYR	ILE	MET	ALA	ASP	LYS	PRO	LYS	ASN	GLY
ATC	AAA	GTT	AAC	TTC	AAA	ATT	AGA	CAC	AAC	ATT	AAA	GAT	GGA	AGC	GTT	CAA	TTA	GCA	GAC
ILE	LYS	VAL	ASN	PHE	LYS	ILE	ARG	HIS	ASN	ILE	LYS	ASP	GLY	SER	VAL	GLN	LEU	ALA	ASP
CAT	TAT	CAA	CAA	AAT	ACT	CCA	ATT	GGC	GAT	GGC	CCT	GTC	CTT	TTA	CCA	GAC	AAC	CAT	TAC
HIS	TYR	GLN	GLN	ASN	THR	PRO	ILE	GLY	ASP	GLY	PRO	VAL	LEU	LEU	PRO	ASP	ASN	HIS	TYR
CTG	TCC	ACG	CAA	TCT	GCC	CTT	TCC	AAA	GAT	CCC	AAC	GAA	AAG	AGA	GAT	CAC	ATG	ATC	CTT
LEU	SER	THR	GLN	SER	ALA	LEU	SER	LYS	ASP	PRO	ASN	GLU	LYS	ARG	ASP	HIS	MET	ILE	LEU
CTT	GAG	TTT	GTA	ACA	GCT	GCT	GGG	ATT	ACA	CAT	GGC	ATG	GAT	GAA	CTA	TAC	AAA	TAA	
LEU	GLU	PHE	VAL	THR	ALA	ALA	GLY	ILE	THR	HIS	GLY	MET	ASP	GLU	LEU	TYR	LYS		

ATGTCCAGACTTCCAATTGACACTAAAGGGATCCGAATTC – 3'

Fig. 3

DNA and predicted primary amino acid sequence of F64L-GFP (Hind3 – EcoR1 fragment).

5' - AAGCTTT

ATG	AGT	AAA	GGA	GAA	GAA	CTT	TTC	ACT	GGA	GTT	GTC	CCA	ATT	CTT	GTT	GAA	TTA	GAT	GGC
MET	SER	LYS	GLY	GLU	GLU	LEU	PHE	THR	GLY	VAL	VAL	PRO	ILE	LEU	VAL	GLU	LEU	ASP	GLY
GAT	GTT	AAT	GGG	CAA	AAA	TTC	TCT	GTT	AGT	GGA	GAG	GGT	GAA	GGT	GAT	GCA	ACA	TAC	GGA
ASP	VAL	ASN	GLY	GLN	LYS	PHE	SER	VAL	SER	GLY	GLU	GLY	GLU	GLY	ASP	ALA	THR	TYR	GLY
AAA	CTT	ACC	CTT	AAA	TTT	ATT	TGC	ACT	ACT	GGG	AAG	CTA	CCT	GTT	CCA	TGG	CCA	ACG	CTT
LYS	LEU	THR	LEU	LYS	PHE	ILE	CYS	THR	THR	GLY	LYS	LEU	PRO	VAL	PRO	TRP	PRO	THR	LEU
GTC	ACT	ACT	CTC	TCT	TAT	GGT	GTT	CAA	TGC	TTT	TCT	AGA	TAC	CCA	GAT	CAT	ATG	AAA	CAG
VAL	THR	THR	LEU	SER	TYR	GLY	VAL	GLN	CYS	PHE	SER	ARG	TYR	PRO	ASP	HIS	MET	LYS	GLN
CAT	GAC	TTT	TTC	AAG	AGT	GCC	ATG	CCC	GAA	GGT	TAT	GTA	CAG	GAA	AGA	ACT	ATA	TTT	TAC
HIS	ASP	PHE	PHE	LYS	SER	ALA	MET	PRO	GLU	GLY	TYR	VAL	GLN	GLU	ARG	THR	ILE	PHE	TYR
AAA	GAT	GAC	GGG	AAC	TAC	AAG	ACA	CGT	GCT	GAA	GTC	AAG	TTT	GAA	GGT	GAT	ACC	CTT	GTT
LYS	ASP	ASP	GLY	ASN	TYR	LYS	THR	ARG	ALA	GLU	VAL	LYS	PHE	GLU	GLY	ASP	THR	LEU	VAL
AAT	AGA	ATC	GAG	TTA	AAA	GGT	ATT	GAT	TTT	AAA	GAA	GAT	GGA	AAC	ATT	CTT	GGA	CAC	AAA
ASN	ARG	ILE	GLU	LEU	LYS	GLY	ILE	ASP	PHE	LYS	GLU	ASP	GLY	ASN	ILE	LEU	GLY	HIS	LYS
ATG	GAA	TAC	AAT	TAT	AAC	TCA	CAT	AAT	GTA	TAC	ATC	ATG	GCA	GAC	AAA	CCA	AAG	AAT	GGC
MET	GLU	TYR	ASN	TYR	ASN	SER	HIS	ASN	VAL	TYR	ILE	MET	ALA	ASP	LYS	PRO	LYS	ASN	GLY
ATC	AAA	GTT	AAC	TTC	AAA	ATT	AGA	CAC	AAC	ATT	AAA	GAT	GGA	AGC	GTT	CAA	TTA	GCA	GAC
ILE	LYS	VAL	ASN	PHE	LYS	ILE	ARG	HIS	ASN	ILE	LYS	ASP	GLY	SER	VAL	GLN	LEU	ALA	ASP
CAT	TAT	CAA	CAA	AAT	ACT	CCA	ATT	GGC	GAT	GGC	CCT	GTC	CTT	TTA	CCA	GAC	AAC	CAT	TAC
HIS	TYR	GLN	GLN	ASN	THR	PRO	ILE	GLY	ASP	GLY	PRO	VAL	LEU	LEU	PRO	ASP	ASN	HIS	TYR
CTG	TCC	ACG	CAA	TCT	GCC	CTT	TCC	AAA	GAT	CCC	AAC	GAA	AAG	AGA	GAT	CAC	ATG	ATC	CTT
LEU	SER	THR	GLN	SER	ALA	LEU	SER	LYS	ASP	PRO	ASN	GLU	LYS	ARG	ASP	HIS	MET	ILE	LEU
CTT	GAG	TTT	GTA	ACA	GCT	GCT	GGG	ATT	ACA	CAT	GGC	ATG	GAT	GAA	CTA	TAC	AAA	TAA	
LEU	GLU	PHE	VAL	THR	ALA	ALA	GLY	ILE	THR	HIS	GLY	MET	ASP	GLU	LEU	TYR	LYS		

ATGTCCAGACTTCCAATTGACACTAAAGGGATCCGAATTC – 3'

Fig. 4

DNA and predicted primary amino acid sequence of F64L-S65T-GFP (Hind3 – EcoR1 fragment).

5' - AAGCTTT

ATG	AGT	AAA	GGA	GAA	GAA	CTT	TTC	ACT	GGA	GTT	GTC	CCA	ATT	CTT	GTT	GAA	TTA	GAT	GGC
MET	SER	LYS	GLY	GLU	GLU	LEU	PHE	THR	GLY	VAL	VAL	PRO	ILE	LEU	VAL	GLU	LEU	ASP	GLY
GAT	GTT	AAT	GGG	CAA	AAA	TTC	TCT	GTT	AGT	GGA	GAG	GGT	GAA	GGT	GAT	GCA	ACA	TAC	GGA
ASP	VAL	ASN	GLY	GLN	LYS	PHE	SER	VAL	SER	GLY	GLU	GLY	GLU	GLY	ASP	ALA	THR	TYR	GLY
AAA	CTT	ACC	CTT	AAA	TTT	ATT	TGC	ACT	ACT	GGG	AAG	CTA	CCT	GTT	CCA	TGG	CCA	ACG	CTT
LYS	LEU	THR	LEU	LYS	PHE	ILE	CYS	THR	THR	GLY	LYS	LEU	PRO	VAL	PRO	TRP	PRO	THR	LEU
GTC	ACT	ACT	CTC	ACT	TAT	GGT	GTT	CAA	TGC	TTT	TCT	AGA	TAC	CCA	GAT	CAT	ATG	AAA	CAG
VAL	THR	THR	LEU	THR	TYR	GLY	VAL	GLN	CYS	PHE	SER	ARG	TYR	PRO	ASP	HIS	MET	LYS	GLN
CAT	GAC	TTT	TTC	AAG	AGT	GCC	ATG	CCC	GAA	GGT	TAT	GTA	CAG	GAA	AGA	ACT	ATA	TTT	TAC
HIS	ASP	PHE	PHE	LYS	SER	ALA	MET	PRO	GLU	GLY	TYR	VAL	GLN	GLU	ARG	THR	ILE	PHE	TYR
AAA	GAT	GAC	GGG	AAC	TAC	AAG	ACA	CGT	GCT	GAA	GTC	AAG	TTT	GAA	GGT	GAT	ACC	CTT	GTT
LYS	ASP	ASP	GLY	ASN	TYR	LYS	THR	ARG	ALA	GLU	VAL	LYS	PHE	GLU	GLY	ASP	THR	LEU	VAL
AAT	AGA	ATC	GAG	TTA	AAA	GGT	ATT	GAT	TTT	AAA	GAA	GAT	GGA	AAC	ATT	CTT	GGA	CAC	AAA
ASN	ARG	ILE	GLU	LEU	LYS	GLY	ILE	ASP	PHE	LYS	GLU	ASP	GLY	ASN	ILE	LEU	GLY	HIS	LYS
ATG	GAA	TAC	AAT	TAT	AAC	TCA	CAT	AAT	GTA	TAC	ATC	ATG	GCA	GAC	AAA	CCA	AAG	AAT	GGC
MET	GLU	TYR	ASN	TYR	ASN	SER	HIS	ASN	VAL	TYR	ILE	MET	ALA	ASP	LYS	PRO	LYS	ASN	GLY
ATC	AAA	GTT	AAC	TTC	AAA	ATT	AGA	CAC	AAC	ATT	AAA	GAT	GGA	AGC	GTT	CAA	TTA	GCA	GAC
ILE	LYS	VAL	ASN	PHE	LYS	ILE	ARG	HIS	ASN	ILE	LYS	ASP	GLY	SER	VAL	GLN	LEU	ALA	ASP
CAT	TAT	CAA	CAA	AAT	ACT	CCA	ATT	GGC	GAT	GGC	CCT	GTC	CTT	TTA	CCA	GAC	AAC	CAT	TAC
HIS	TYR	GLN	GLN	ASN	THR	PRO	ILE	GLY	ASP	GLY	PRO	VAL	LEU	LEU	PRO	ASP	ASN	HIS	TYR
CTG	TCC	ACG	CAA	TCT	GCC	CTT	TCC	AAA	GAT	CCC	AAC	GAA	AAG	AGA	GAT	CAC	ATG	ATC	CTT
LEU	SER	THR	GLN	SER	ALA	LEU	SER	LYS	ASP	PRO	ASN	GLU	LYS	ARG	ASP	HIS	MET	ILE	LEU
CTT	GAG	TTT	GTA	ACA	GCT	GCT	GGG	ATT	ACA	CAT	GGC	ATG	GAT	GAA	CTA	TAC	AAA	TAA	
LEU	GLU	PHE	VAL	THR	ALA	ALA	GLY	ILE	THR	HIS	GLY	MET	ASP	GLU	LEU	TYR	LYS		

ATGTCCAGACTTCCAATTGACACTAAAGGGATCCGAATTC – 3'

Fig. 5

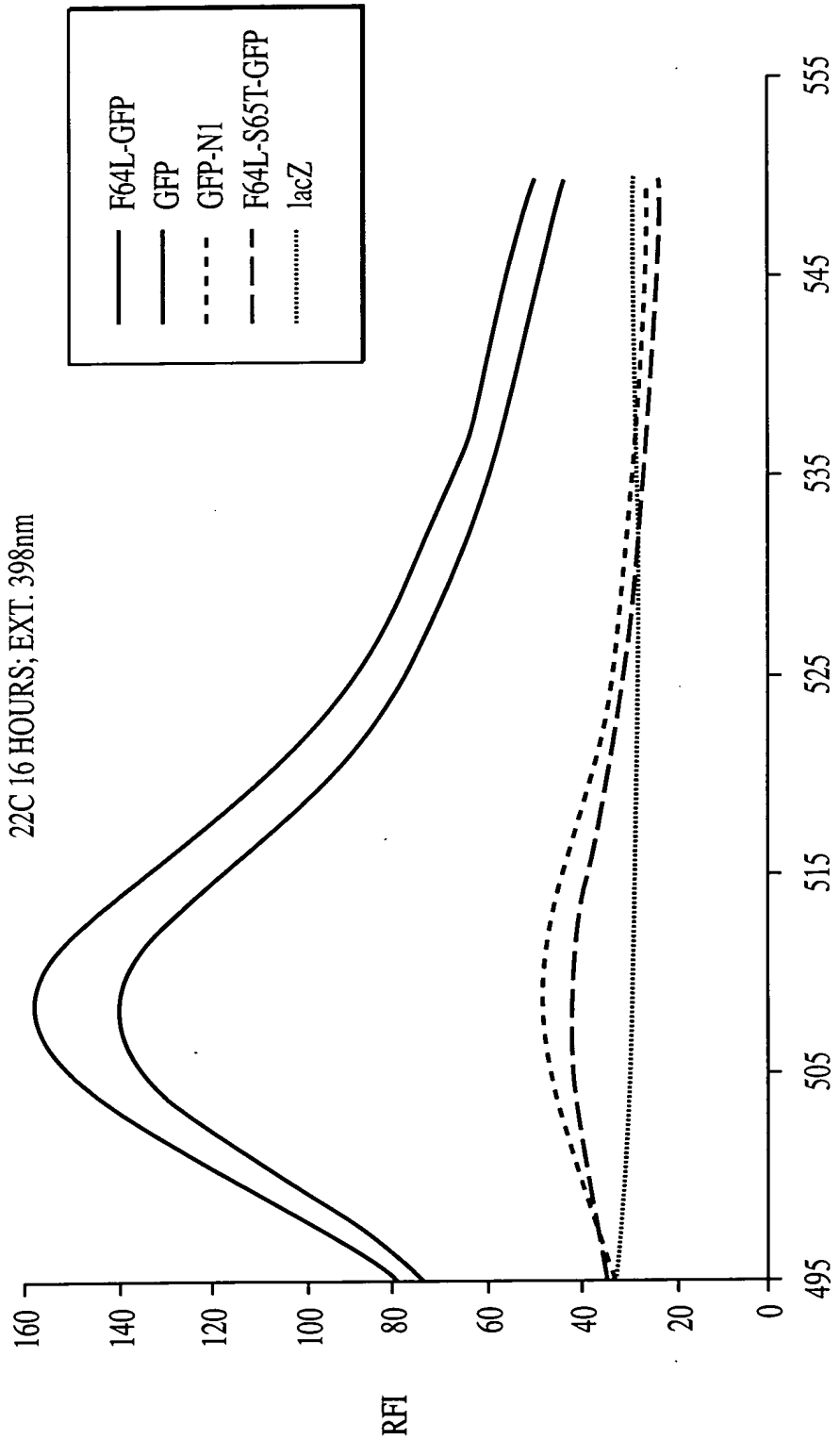
22C 16 HOURS; EXT. 398nm
F64L-GFP
GFP
GFP-N1
F64L-S65T-GFP
lacZ
RFI
160
140
120
100
80
60
40
20
0
495
505
515
525
535
545
555
nm

FIG. 6A

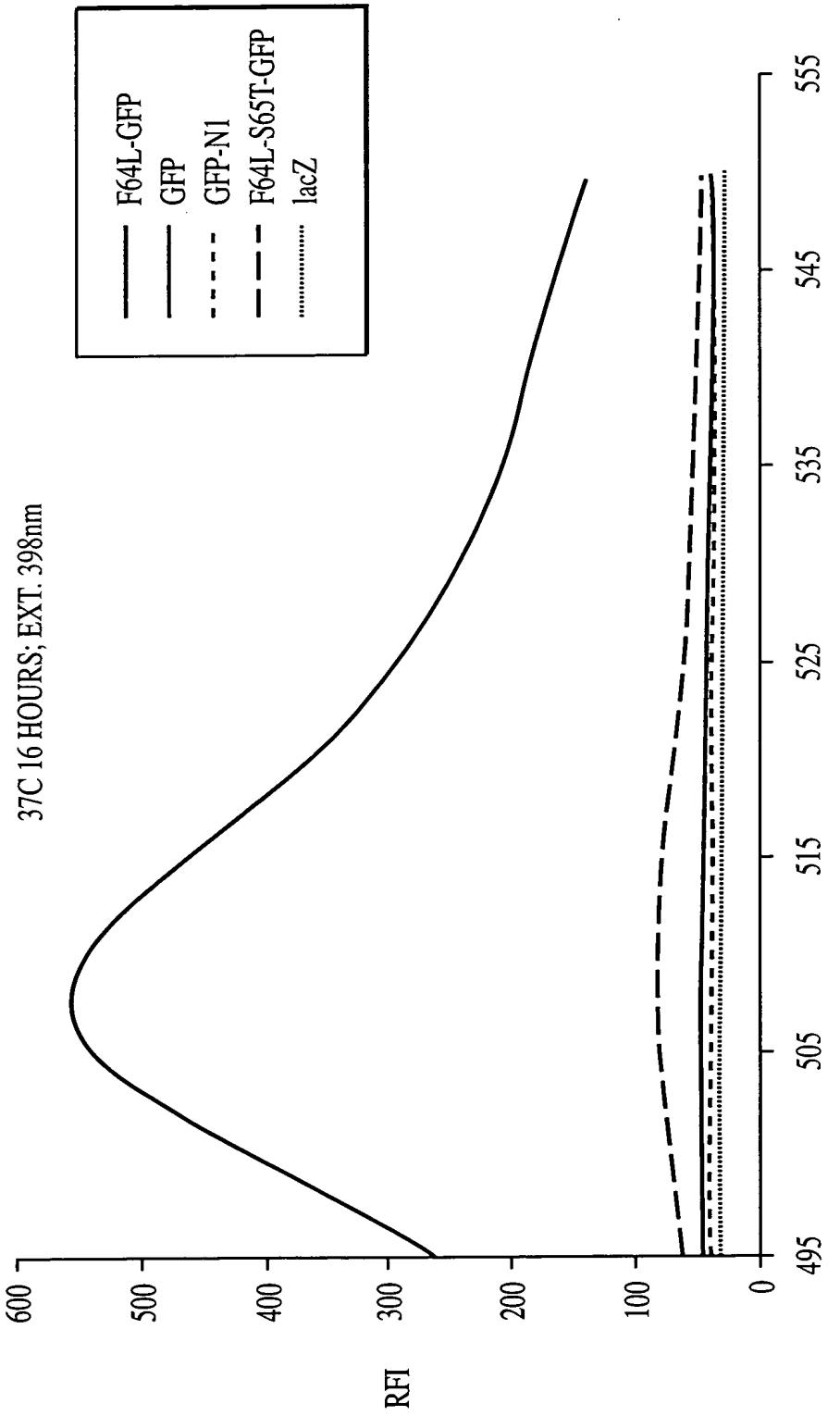
F64L-GFP
GFP
GFP-N1
F64L-S65T-GFP
lacZ
37C 16 HOURS; EXT. 398nm
RFI
600
500
400
300
200
100
0
495
505
515
525
535
545
555
nm

FIG. 6B

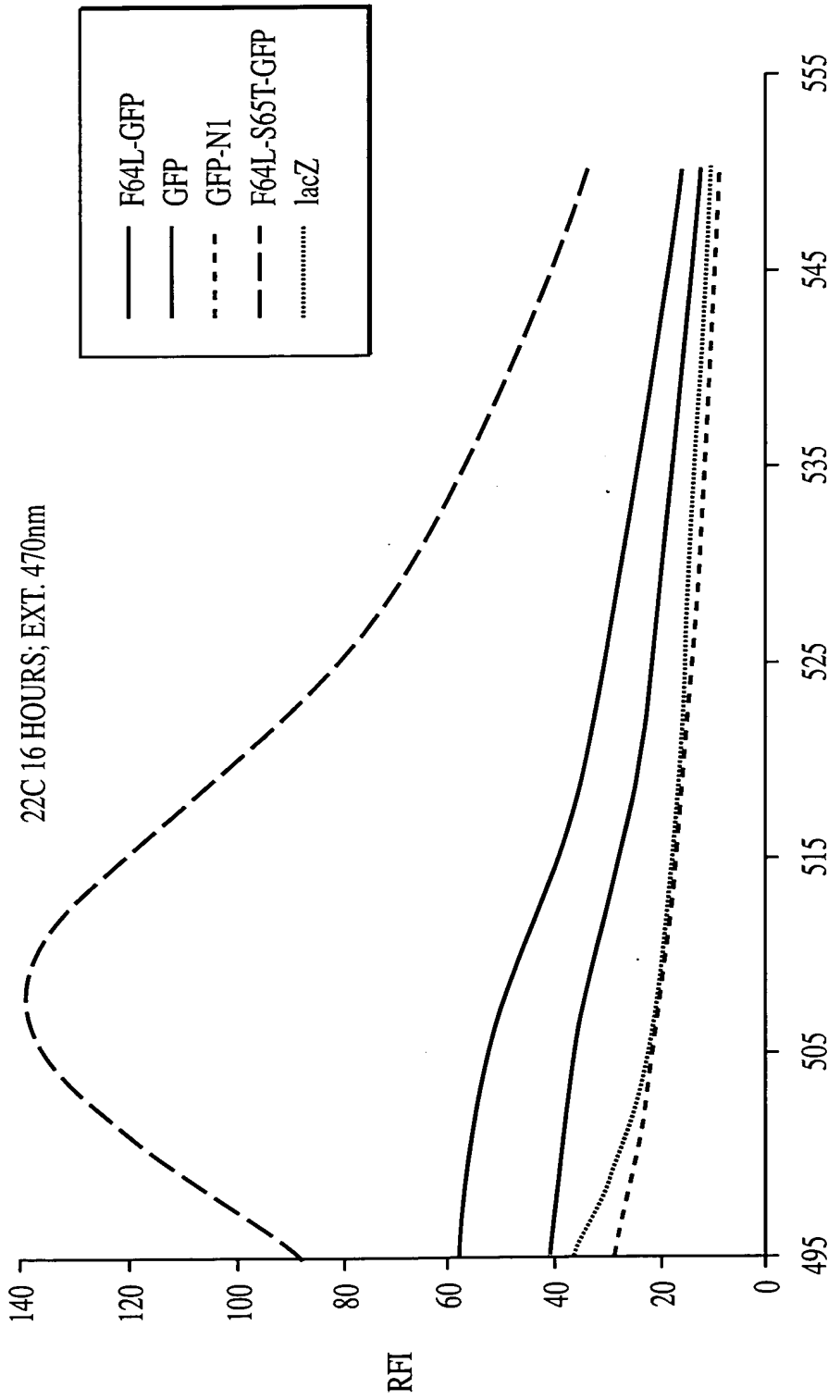
F64L-GFP
GFP
GFP-N1
F64L-S65T-GFP
lacZ
22C 16 HOURS; EXT. 470nm
RFI
140
120
100
80
60
40
20
0
495
505
515
525
535
545
555
nm

FIG. 6C

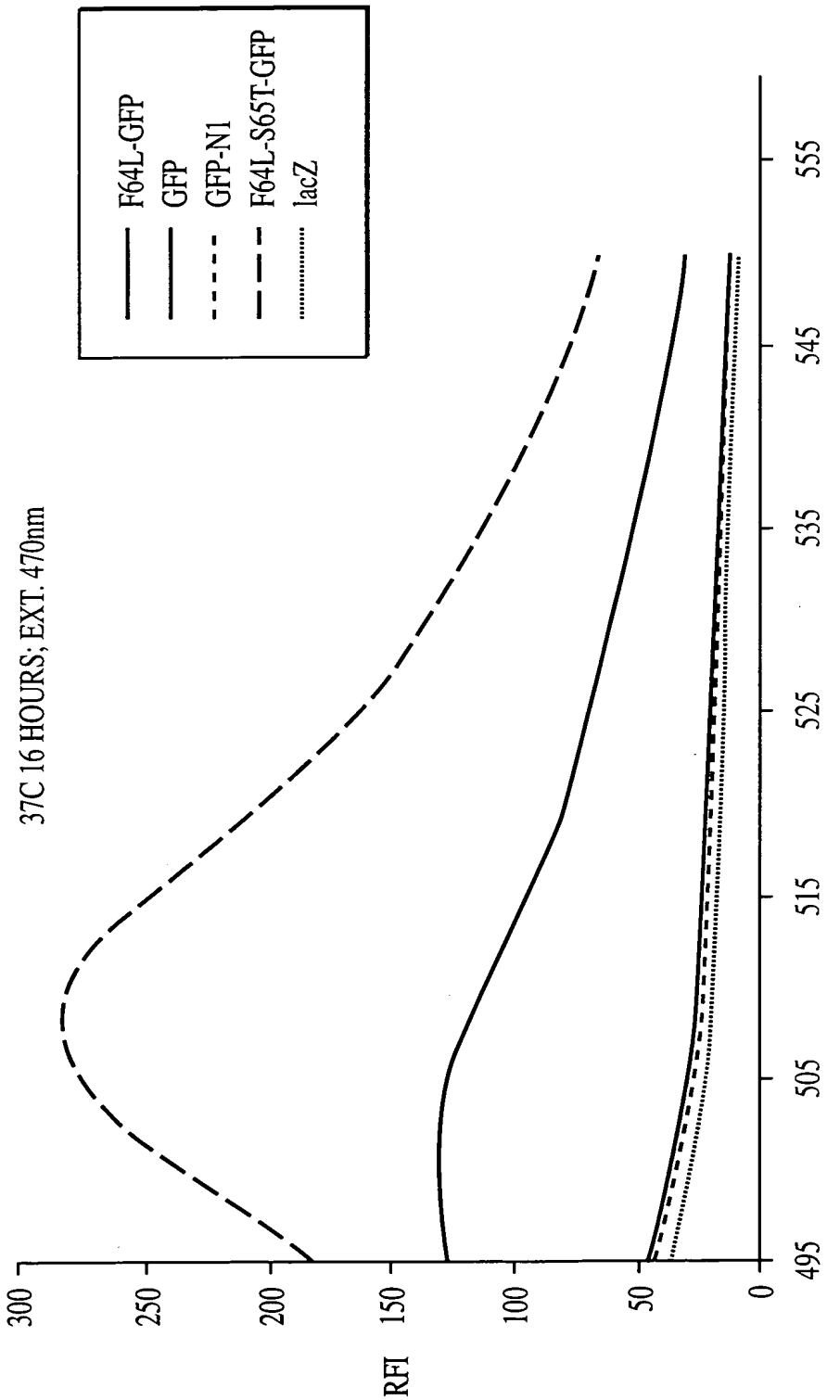
37C 16 HOURS; EXT. 470nm
F64L-GFP
GFP
GFP-N1
F64L-S65T-GFP
lacZ
RFI
300
250
200
150
100
50
0
495
505
515
525
535
545
555
nm

FIG. 6D

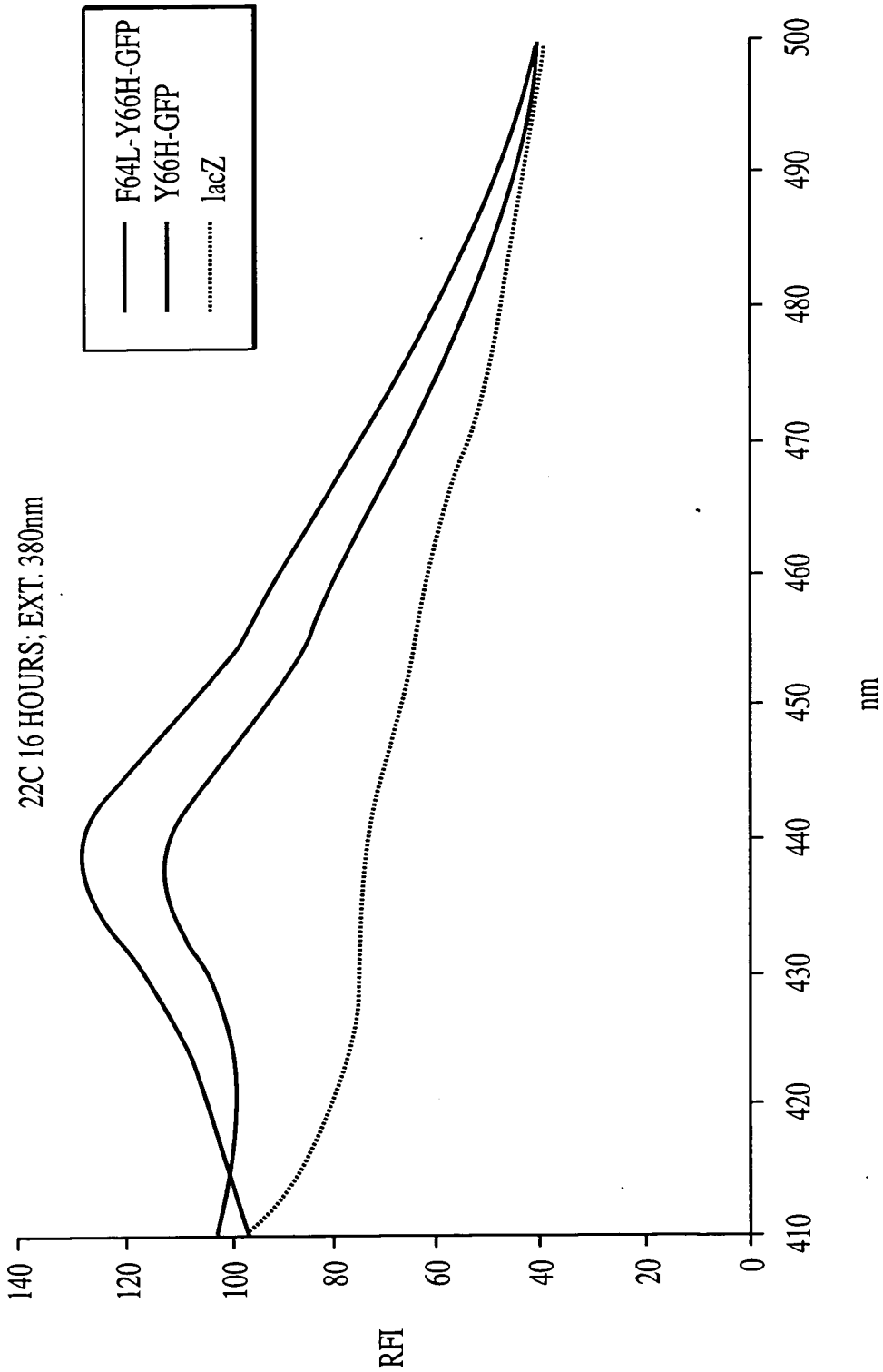
F64L-Y66H-GFP
Y66H-GFP
lacZ
22C 16 HOURS; EXT. 380nm
RFI
nm
140
120
100
80
60
40
20
0
410
420
430
440
450
460
470
480
490
500

FIG. 6E

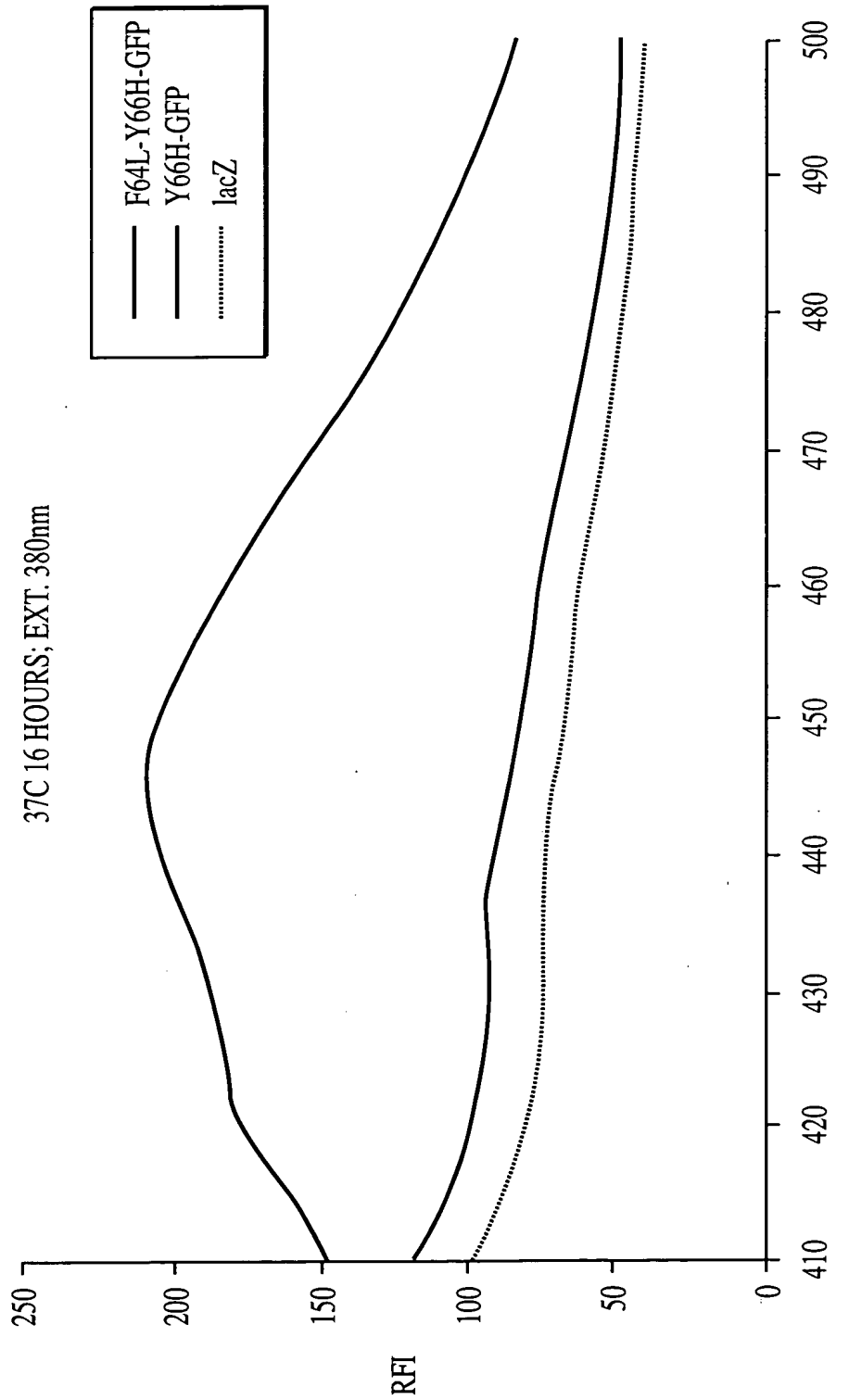
F64L-Y66H-GFP
Y66H-GFP
lacZ
37C 16 HOURS; EXT. 380nm
RFI
250
200
150
100
50
0
410
420
430
440
450
460
470
480
490
500
nm

FIG. 6F